

Current issues in medically assisted reproduction and genetics in Europe: research, clinical practice, ethics, legal issues and policy[†]

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Submitted on April 2, 2014; resubmitted on April 2, 2014; accepted on May 2, 2014

STUDY QUESTION: How has the interface between genetics and assisted reproduction technology (ART) evolved since 2005?

SUMMARY ANSWER: The interface between ART and genetics has become more entwined as we increase our understanding about the genetics of infertility and we are able to perform more comprehensive genetic testing.

WHAT IS KNOWN ALREADY: In March 2005, a group of experts from the European Society of Human Genetics and European Society of Human Reproduction and Embryology met to discuss the interface between genetics and ART and published an extended background paper, recommendations and two Editorials.

STUDY DESIGN, SIZE, DURATION: An interdisciplinary workshop was held, involving representatives of both professional societies and experts from the European Union EuroGentest2 Coordination Action Project.

PARTICIPANTS/MATERIALS, SETTING, METHODS: In March 2012, a group of experts from the European Society of Human Genetics, the European Society of Human Reproduction and Embryology and the EuroGentest2 Coordination Action Project met to discuss developments at the interface between clinical genetics and ART.

[†]ESHRE Pages content is not externally peer reviewed. The manuscript has been approved by the Executive Committees of ESHRE and ESHG.

MAIN RESULTS AND THE ROLE OF CHANCE: As more genetic causes of reproductive failure are now recognized and an increasing number of patients undergo testing of their genome prior to conception, either in regular health care or in the context of direct-to-consumer testing, the need for genetic counselling and PGD may increase. Preimplantation genetic screening (PGS) thus far does not have evidence from RCTs to substantiate that the technique is both effective and efficient. Whole genome sequencing may create greater challenges both in the technological and interpretational domains, and requires further reflection about the ethics of genetic testing in ART and PGD/PGS. Diagnostic laboratories should be reporting their results according to internationally accepted accreditation standards (ISO 15189). Further studies are needed in order to address issues related to the impact of ART on epigenetic reprogramming of the early embryo.

LIMITATIONS, REASONS FOR CAUTION: The legal landscape regarding assisted reproduction is evolving, but still remains very heterogeneous and often contradictory. The lack of legal harmonization and uneven access to infertility treatment and PGD/PGS fosters considerable cross-border reproductive care in Europe, and beyond.

WIDER IMPLICATIONS OF THE FINDINGS: This continually evolving field requires communication between the clinical genetics and IVF teams and patients to ensure that they are fully informed and can make well-considered choices.

STUDY FUNDING/COMPETING INTEREST(S): Funding was received from ESHRE, ESHG and EuroGentest2 European Union Coordination Action project (FP7 – HEALTH-F4-2010-26146) to support attendance at this meeting.

Key words: assisted reproduction technology / European Society of Human Genetics / European Society of Human Reproduction and Embryology / reproductive genetics / IVF

Introduction

In 2004, the Public and Professional Policy Committee (PPPC) of the European Society of Human Genetics (ESHG) felt the need for professional recommendations on how to use assisted reproduction technology (ART) safely and reliably from the genetic point of view, as well as issuing guidelines on acceptable (genetic) goals of ART treatment and its prioritization in the European healthcare systems. It was thus decided to approach the European Society of Human Reproduction and Embryology (ESHRE) to undertake such work together. After a number of preparatory get-togethers of a working group, a joint ESHG/ESHRE meeting was held in Seville, Spain, from 31 March to 1 April 2005, and a paper summarizing the meeting was published (Soini *et al.*, 2006).

In 2012, an expert group of ESHRE and ESHG representatives met again to assess the changes that had occurred in the field for the past 7 years and to update the common background document, wherever needed. This commentary presents a summary of the meeting which is also presented in full (Harper *et al.*, 2013).

European Directives

Recent European Union (EU) directives have had a significant impact on ART. In particular, the EU Tissue and Cells Directive (EUTCD; currently under revision) (EC-Dir23, 2004) and the supplementing technical directives 2006/17/EC (EC-Dir17, 2006) and 2006/86/EC (EC-Dir86, 2006; Willemen *et al.*, 2012) have led to new safety and quality standards for clinical and laboratory procedures performed within IVF. Most European countries already transposed them into their respective national legislations, thus regulating procurement, testing, processing, storage, distribution and import/export of reproductive cells and tissues. Moreover, the EU Directive 98/79/EC (EC-Dir98-IVD, 1998) on *in vitro* diagnostic medical devices, known as the 'IVD Directive' is also currently under revision and may have a significant effect on the field of genetic testing and its interface with ART.

Cross-Border Reproductive Care

Cross-border reproductive care (CBRC) refers to the care of patients who cross borders in search of reproductive treatment. Recent European data from the first international European study published by the CBRC ESHRE Taskforce (Shenfield *et al.*, 2010) confirmed that the main reasons for travelling were legal restrictions based on prohibition of the technique *per se*, or because of inaccessibility due to the characteristics of the patients (like age, sexual orientation or civil status). It also highlighted that people tend to travel to the nearest country where the required technique is available, such as Germans seeking oocyte donation in the Czech Republic, or French lesbian couples donor insemination in Belgium.

Concerned about the joint professional responsibility of all involved in ART, the CBRC ESHRE Taskforce also published a Good Practice Guide for CRBC, for patients and collaborators involved in the child project, including gamete donors and surrogates (Shenfield *et al.*, 2011).

Other challenges are also looming, as oocyte banking is becoming much more efficient thanks to vitrification (Cobo *et al.*, 2008). This means that eggs have started to cross borders, making it more difficult to check the conditions of gamete donation.

Genetics of Female Infertility

For the more common conditions observed in the infertile female, such as polycystic ovary syndrome (PCOS) (Koika *et al.*, 2012), endometriosis (Rahmioglu *et al.*, 2012), and certain anomalies of the female reproductive system, no diagnostic genetic test is available as these conditions are multifactorial but there is an increase in the number of genetic tests being offered for female infertility.

Genetic testing of females with ovarian insufficiency and amenorrhoea should consist of a chromosomal analysis and fragile X mental retardation-1 gene (FMR1) 'expansion' CGG(n) testing (Foresta *et al.*, 2002). Other more specific tests looking for the BPES (blepharophimosis-ptosis-epicanthus inversus) syndrome caused by FOXL2 mutations (Beysen *et al.*, 2009), galactosemia (GLAT mutations) (Calderon *et al.*,

2007) or less commonly POLG mutations (Tong *et al.*, 2010) associated with a mitochondrial disease should be performed if clinically indicated. Rarer causes of female infertility, such as those due to mutations in the FSH-receptor (FSHR) or the LH/CG receptor (LHCGR), should also be kept in mind and investigated whenever necessary (De Vos *et al.*, 2010; Persani *et al.*, 2010). Other rare hereditary conditions, such as Kallmann syndrome (Cadman *et al.*, 2007), androgen insensitivity (Hughes *et al.*, 2012) and adrenal hyperplasia may be diagnosed as causes of female infertility (ESHRE-Capri, 2008). In cases of recurrent miscarriages (at least three times) (Rull *et al.*, 2012), and for examining balanced translocations and/or other structural anomalies, chromosome analysis should be performed (ESHRE-Capri, 2008). Inherited thrombophilia testing requires standardization and further meta-analyses in order to substantiate its clinical utility (McNamee *et al.*, 2012).

New technologies, such as array comparative genomic hybridization (aCGH), whole exome sequencing (WES) or whole genome sequencing (WGS) will likely enable identification of additional genes that play an important role in female reproductive failure (Sykiotis *et al.*, 2010).

Genetic Aspects of Male Infertility

It has been estimated that roughly one quarter of patients with azoospermia (AZ) and severe oligozoospermia undergo genetic testing (Stahl and Schlegel, 2012). The first examination of choice is karyotyping for common chromosomal aberrations, possibly followed by specific molecular tests. Interestingly, the lower the sperm count, the more chromosomal aberrations are found (Dul *et al.*, 2012).

Gonosomal aberrations are mainly represented by Klinefelter syndrome (Maiburg *et al.*, 2012) (47,XXY, including its various mosaic formulae) with a rather heterogeneous clinical presentation. Pathogenic mutations or variants in the cystic fibrosis transmembrane receptor gene (CFTR) have been implicated in male infertility (Xu *et al.*, 2007) and are associated with obstructive AZ due to congenital bilateral absence of the vas deferens (CBAVD) (Yu *et al.*, 2012).

Various Y chromosome microdeletions are predominantly found in non-obstructive AZ or severe oligospermia (Simoni *et al.*, 2008). Molecular genetic diagnosis of such microdeletions is useful and feasible with a simple and robust test (Simoni *et al.*, 2004). The association to sperm counts is proportional, in that AZ men have a higher prevalence of microdeletions than oligospermic patients. Testing of AZFI microdeletions has a prognostic impact for sperm extraction, since no sperm can be retrieved in AZFI a and AZFI b, while there is a fair chance that viable sperm could be retrieved in AZFI c (Navarro-Costa *et al.*, 2010; Patrat *et al.*, 2010; Rozen *et al.*, 2012).

Alterations in the GnRH gene (GNRH1), implicated in impaired neuronal migration (Kim *et al.*, 2008), were found in congenital hypogonadotrophic hypogonadism (Sato, 2012). Mutations in the androgen receptor (AR) gene (Ferlin *et al.*, 2006) are found in ~1% of patients with AZ or severe oligozoospermia. It should be noted that examination of the entire AR gene is needed.

New technology, such as single nucleotide polymorphism (SNP) variants (Tuttelmann *et al.*, 2007) and genome-wide association studies (GWAS) (Kosova *et al.*, 2012), has provided evidence that there are multiple SNPs significantly associated with decreased sperm counts (Hu *et al.*, 2012). At the moment there is no routine 'diagnostic' indication

for WES-/WGS-based approaches in male or female infertility (Pastuszak and Lamb, 2012).

Counselling Within the Reproductive Medicine and Genetic Contexts

Genetic counselling is a communication process that involves discussing the problems associated with a genetic condition (ASHG-GC, 1975; Resta *et al.*, 2006). A typical genetic counselling consultation encompasses the familial, scientific, psychological and social aspects of being at risk or being affected. Key concepts in genetic counselling practice are relevant information exchange, the presentation of choices relevant to the patient and exploration of patient values and beliefs. In traditional genetic counselling (of fertile prospective parents), a non-directive approach is considered to be of paramount importance.

Where genetic risks are related to the cause of infertility, genetic counselling is always required. The cause itself is explained and the implications for the person's close relatives (e.g. siblings).

Some couples opt for ART treatment because the fetus is at high risk for a genetic condition and they wish to avoid termination of pregnancy. There are several options available to couples that wish to avoid having an affected child; including having no children, having no genetic testing, having prenatal diagnosis, PGD, using donor gametes, or adoption. For PGD, genetic counselling should be provided by trained professionals.

PGD and Preimplantation Genetic Screening

PGD is a diagnostic test used to select genetically or chromosomally normal embryos for patients at high risk of transmitting a specific abnormality to their children. Even though these patients will often be fertile, they have to undergo IVF/ICSI to generate embryos *in vitro*, which will be biopsied and these cells will subsequently undergo genetic testing. The disease or chromosome abnormality needs to be previously identified, so that specific (targeted) genetic testing could be performed. Embryos that are free from the disease tested will be transferred into the uterus.

Preimplantation genetic screening (PGS) is an adjunct to IVF and is used to aid embryo selection for certain groups of patients including those with advanced maternal age, repeated IVF failure, repeated miscarriage with normal karyotypes in the parents and severe male factor.

For both PGD and PGS, genetic testing can be performed on the polar bodies, blastomeres from cleavage-stage embryos or trophectoderm cells from the blastocyst (Harton *et al.*, 2011d).

Historically, two main techniques have been used for the genetic analysis in PGD (Harper and Sengupta, 2012): a PCR-based test for single gene defects (Harton *et al.*, 2011b) and fluorescence *in situ* hybridization (FISH) to examine chromosomes (for PGD of translocations and PGS) (Harton *et al.*, 2011c). More recently, arrays have replaced FISH to examine chromosomes (Harper and Harton, 2010).

The use of oligo-/SNP arrays (Handyside *et al.*, 2010; Brezina *et al.*, 2011; Treff *et al.*, 2011) and WES/WGS analyses will allow a substantial increase in the amount of genetic information that will become available from each embryo. These rapid technological developments will

necessitate development of novel guidelines, interpretation algorithms and ethical frameworks.

PGS has increasingly been used in the past decade but RCTs have failed to show its effectiveness. PGS RCTs using FISH have been shown to significantly reduce the live birth rate (Harper et al., 2010a,b; Mastenbroek et al., 2011). ESHRE conducted a proof-of-principle study to validate the use of aCGH for aneuploidy screening (Geraedts et al., 2011) and has initiated a multi-centre polar body RCT using aCGH for advanced maternal age (ongoing at the time of publication). Three recent RCTs reported on using new technology for PGS but they have a small sample size and were conducted on good prognosis patients, not the patients that PGS would be indicated for (Scott et al., 2012; Yang et al., 2012; Scott et al., 2013).

The ESHRE PGD Consortium has analysed over 35 000 cycles of PGD/PGS since 1997 (Goossens et al., 2012; Harper et al., 2012a). The Consortium has recently produced four guidelines: the organization of a PGD/PGS centre, amplification-based PGD, FISH-based PGD and embryology as it relates to PGD and PGS (Harton et al., 2011a,b,c,d). The Consortium has also produced a guide to PGD laboratory accreditation (Harper et al., 2010a,b).

Genomic Variation in Early Human Development and Related Diagnostic Techniques

It is by now well established that chromosomal abnormalities are inherent to human embryos (Delhanty et al., 1993; Munne et al., 1994; Iwarson et al., 1999; Munne et al., 2004; Baart et al., 2006; Hodes-Wertz et al., 2012). Meta-analyses reviewing 36 studies, in which all blastomeres of cleavage-stage embryos have been analysed by at least eight FISH probes, show that only 22% of embryos are euploid (van Echten-Arends et al., 2011), with an increase up to 45% in blastocysts (Fragouli and Wells, 2011). Considering the mitotic error rate during the cleavage stage, analysis of single blastomeres will not provide insight in the genomic constitution of the other cells, nor in the developmental potential of the embryo (Vanneste et al., 2009; Vanneste et al., 2012).

Single cell WES/WGS sequencing for PGD will provide the most in-depth view of the human genome at early stages of human development (Navin et al., 2011; Spencer and Palmarini, 2012). It is important to assess the evolutionary, medical, ethical and legal consequences of these novel technologies in both clinical and community genetics and assisted human reproduction (Dondorp and de Wert, 2013; Pennings and Mertes, 2012).

Accreditation of Laboratories in the Field of Reproductive Genetics

The appropriate accreditation standard for medical diagnostic laboratories is ISO 15189 (ISO 15189, 2012). Accreditation to this (or an equivalent) is considered as the single most effective route to comprehensive laboratory quality assurance as stipulated by the Organisation for Economic Cooperation and Development (OECD) 'Guidelines for Quality Assurance in Genetic testing' (OECD-QA, 2007). Validation is a fundamental requirement of ISO 15189: 'The methods and procedures

selected for use shall be evaluated and found to give satisfactory results before being used for medical examinations'.

External quality assessment (EQA) and other methods of inter-laboratory comparison are a formal requirement of ISO 15189. The ESHRE PGD Consortium set up a network of partner organizations: (i) PGD-specific EQA schemes are now available, (ii) training workshops addressing the accreditation of PGD laboratories are held regularly, (iii) the Consortium has formally recommended that all PGD laboratories should be accredited or working actively towards accreditation (ISO 15189) and (iv) a guidance document has been published (Harper et al., 2010b).

Direct-to-Consumer Genetic Testing at the Interface of Genetics and Reproduction

Genetic tests are usually performed in a clinical (medical) genetics centre with access to counselling (CoE-GenetHealthcare, 2010). In contrast, within the last several years primarily commercial entities have been increasingly advertising and selling genetic tests either direct-to-consumers (DTC) or providing them to their customers in a 'DTC-through-physician' (i.e. via a nonspecialist) manner (Hunter et al., 2008; EASC/FEAM report, 2012). DTC genetic testing can be defined as the advertising and selling or (free) provision of genetic tests directly to consumers.

The range of DTC genetic tests available is broad, including carrier tests for (i) Mendelian genetic disorders (rare diseases), (ii) 'life style'-related genetic traits, (iii) pharmacogenomics, (iv) non-invasive prenatal testing (v) paternity, (vi) 'romantic relationship' testing, (vii) genomic risk profiles for many conditions, or (viii) 'recreational' ancestry or genealogical tests (Howard and Borry, 2011). These different types of tests bring different practical and ethical concerns (Borry and Howard, 2008).

It is expected that DTC companies will be offering WES/WGS in the near future. WGS poses ethical problems, because it basically provides complete information about an individual genome (Su et al., 2011). This means that, potentially, every single trait or disorder ever associated in the past (or ever to be associated with in the future) with a SNP or other genomic alteration could be identified.

The commercial offer of carrier testing through the internet creates various challenges. The large number of disorders that are included in most of the screening panels contrasts with the limited amount of disorders that are usually suggested to be screened and customers are referred to their individual physicians for further interpretation of test results. The majority of these companies 'disclaim' any responsibility for the quality of their service (Borry et al., 2009; Borry et al., 2010; Howard et al., 2011).

Epigenetic Effects Related to ART

One of the recurrent questions in ART is focused on the issue as to how much this medical technology could affect the epigenome of human embryos produced *in vitro*. The epigenome comprises the complete set of non-covalent modifications onto the genetic material of a cell or an organism (Yuan, 2012). These epigenetic marks often affect transcriptional activity and control developmental plasticity of cells, including

cell-type-specific gene expression patterns (Jablonka and Raz, 2009; van Montfort *et al.*, 2012).

Studies on animal models have established that environmental factors, such as ovulation induction, culture medium composition and/or embryo manipulation, might affect the epigenome and impact on the conceptus, including neonatal birthweight. Animal models such as mouse and cow, commonly suffer from the so-called 'large-offspring syndrome' (Khosla *et al.*, 2001; Young *et al.*, 2001; Velker *et al.*, 2012). In humans, more studies are needed to corroborate initial discrepant observations with regard to low birthweight babies following ART (Dumoulin *et al.*, 2010).

Genomic imprinting is a process through which alleles of given genes are expressed in a parent-of-origin-specific manner. Genes that are subject to imprinting often play key roles in embryonic development and behaviour. In humans, several defects in imprinted genes are linked to syndromes such as Beckwith–Wiedemann (MIM #130650), Prader–Willi (MIM #176270), Angelman (MIM #105830) and Silver–Russell (MIM #180860). Increased prevalence of imprinting disorders related to ART has been reported, but most often without proper reference to the primary cause of infertility and methods (ICSI, culture media). A summary of the evidence on imprinting and ART shows that absolute risks appear to be low.

The influence of ART on the status of the epigenome is not yet completely understood (Odom and Segars, 2010). ESHRE understands and is supporting initiatives for RCTs of culture media and their impact on early epigenetic programming in the embryo. This may also be relevant when evaluating imprinting defects after ART.

Epidemiological Aspects: Birth Defects and Population Genetics

ART is associated with a slightly elevated risk of birth defects, multiple pregnancies (leading to pre- and dysmaturity) and may contribute to an increase of the genetic causes of fertility problems in the future. To evaluate the pros- and cons of ART, prospective, large cohort, lifelong, multigenerational and multi-centre follow-up studies would be extremely important (Soini *et al.*, 2006), and this issue needs to be addressed at an international level.

In a systematic review of outcomes after ICSI, eight relevant studies were identified, two studying karyotypes and five reporting malformations (Odom and Segars, 2010). In total, there were 55/1973 (2.8%) abnormal karyotypes in the ICSI group with ejaculated sperm, 0/31 in the ICSI group with epididymal sperm and 5/191 (2.6%) in the ICSI group with testicular sperm. Major malformations were found after ICSI in 543/12 377 (4.4%) in the ejaculated sperm group, 17/533 (3.2%) in the epididymal sperm group and 31/670 (4.6%) in the testicular sperm group. While these show that over 95% of infants do not have these health problems, they have no statistical power to exclude an increase in specific birth defects.

A large registry-based study analysing the birth defects registry in the USA, including 1% of ART in the population and 13,500 infants with birth defects, reported that among singleton births, ART was associated with septal heart defects (adjusted odds ratio (aOR) = 2.1, 95% confidence interval (CI) 1.1–4.0), cleft lip with or without cleft palate (aOR = 2.4, 95% CI 1.2–5.1), oesophageal atresia (aOR = 4.5, 95% CI 1.9–10.5) and anorectal atresia (aOR = 3.7, 95% CI 1.5–9.1) (Reefhuis *et al.*, 2009).

Outcomes of pregnancies after IVF were studied in Sweden over a period of 25 years and revealed a decrease of multiple pregnancies, a decrease of pre-eclampsia and premature rupture of membranes, and an increased risk for cerebral palsy, possibly for attention deficit and hyperactivity disorder, for impaired visual acuity and for childhood cancer, though stressing that these outcomes were generally rare, even after IVF (Finnstrom *et al.*, 2011).

To decide whether or not increased risk exists, and whether or not they are avoidable or acceptable, more interdisciplinary research is needed.

Human Embryonic Stem Cells and Induced Pluripotent Stem Cells: Pitfalls and Promises for Regenerative Medicine and Disease Modelling

Pluripotency is usually defined as the ability of a cell to differentiate into derivatives of the three germ layers. Human embryonic stem cells (hESC) are the best known example of pluripotent cell lines (Ben-David *et al.*, 2012), and are for the largest part derived from the inner cell mass of 5- to 6-day old blastocysts. A major breakthrough in the field was the demonstration that terminally differentiated somatic cells could be re-programmed into a pluripotent state by the induced expression of only four key pluripotency genes (OCT4, KLF4, SOX2 and C-MYC). Many observers consider these induced pluripotent stem cells (iPSC) to be the future replacement of hESC, as they do not carry the negative connotation of embryo research (Tiscornia *et al.*, 2011).

In past years, an increasing body of evidence has accumulated showing that hESC and iPSC suffer from genomic instability that is reminiscent of cancer cells. They quickly acquire trisomies, especially for chromosomes 12 and 17, small recurrent amplifications in chromosome 20 (Spits *et al.*, 2008) and mitochondrial mutations (Van Haute *et al.*, 2013), and their epigenome changes haphazardly (Amps *et al.*, 2011). Further work is still needed to establish optimal culture conditions that prevent or limit this instability. Concurrently, robust and higher throughput screening tests need to be developed in order to assess the genomic/chromosomal stability of stem cells *in vitro*, both for research, diagnostic, and eventually for therapeutic purposes.

Equal Access, Prevention of Infertility, Public Funding of Assisted Reproduction

Equal access to assisted reproduction for those with similar reproductive needs is still not a reality in Europe. Restrictive national legal provisions lead to increased CBRC (Shenfield *et al.*, 2010) and create further barriers and social injustice. The EU and its Member State (MS) national health authorities should enable equal access to ART and PGD, as part of regular health care and favour education about infertility, genetics and reproductive options (Pennings *et al.*, 2008a,b; CoE-PGD-PND, 2011).

The adequate role of genetics in health care systems, including promotion of public awareness on recent advances in genetics and of their impact

on the general population, were comprehensively outlined in the Council of Europe (CoE)- 'Recommendation CM/Rec (2010)11 of the Committee of Ministers to MS on the impact of genetics on the organization of health care services and training of health professional's (CoE-GenetHealthcare, 2010), in the drafting of which several members of ESHG and ESHRE were involved.

The field has a responsibility to contribute to enlarging access to assisted reproduction by efforts to increase effectiveness and reduce costs, especially when services are publicly funded.

Ethical Issues Related to Assisted Reproduction and Reproductive Genetics

It is generally accepted that professionals providing medically assisted reproduction have a responsibility to take account of the welfare of the future child. According to ESHRE's Task Force Ethics & Law, they should refrain from participating in reproduction if there is a high risk that the future child would have a seriously diminished quality of life. Professional co-responsibility for the welfare of the child marks an important difference between the normative framework of medically assisted reproduction and that of traditional genetic counselling, with its emphasis on non-directiveness (De Wert, 1999).

PGD is a field with many circumstances for discussion about specific issues. For instance, PGD for HLA-typing in order to conceive a 'saviour sibling' for a child with a life-threatening disorder (Shenfield, 2005), and PGD for disorders caused by a mitochondrial DNA defect (Bredenoord et al., 2008).

The introduction of new reproductive technologies requires more evidence about their efficacy, safety and cost-effectiveness, as well as long-term follow up of clinical data (Harper and Sengupta, 2012). Innovations should first be tested in preclinical animal and embryo studies for efficacy and safety (Harper et al., 2012; Brown and Harper, 2012). Since this may also require research involving the creation of human embryos, the fact that this type of embryo research is forbidden in many countries (as well as by the European Convention on Human Rights and Biomedicine (Coe-Oviedo, 1997) poses a hurdle for the responsible introduction of new technologies (Dondorp and de Wert, 2011).

Another area at the interface of assisted reproduction and genetics is the selection of gamete donors. As more diseases with a strong genetic component are being identified, and as there are increasing technical capacities to detect them, the question arises whether the present guidelines for genetic donor screening need revision (Daar and Brzyski, 2009). Clearly, a 'zero risk' approach is unrealistic and may lead to false reassurance. Relevant ethical issues not only relate to the interests of the prospective parents and the child to be, but also to those of the donor.

New ethical issues can be expected to arise as a result of the introduction of arrays and WES/WGS in the context of PGD and PGS (Harper and Harton, 2010; Harper and Sengupta, 2012; Hens et al., 2013). A possible future scenario is that one 'universal' genome analysis will routinely be offered to all those seeking assisted reproduction, possibly in combination with preconception testing (Handyside and Xu, 2012). From an ethical point of view, this not only raises concerns about adequate pre-test counselling and informed consent, but also requires a further rethinking of the aims of PGD/PGS (De Wert, 2009). Comprehensive testing of embryos may lead to finding predispositions for late onset

disorders for which no adequate options for treatment or prevention exist. ESHG guidelines stipulate that predictive testing for such disorders should not be done in minors (Borry et al., 2009; ESHG-Minors, 2009). If that is the case, would it be acceptable to bring children into the world with a positive outcome of the same kind of testing? Some of the moral problems of WES/WGS testing-based PGS may be avoided with the alternative strategy of offering preconception screening to prospective parents followed by targeted PGD in case of high risk. Obviously, the ethics of this approach needs further scrutiny (De Wert et al., 2012).

Legal Issues Related to Assisted Reproduction and Reproductive Genetics

Major differences still exist in Europe (CoE-PGD-PND, 2011). PGD is banned in Austria and Switzerland, whereas jurisprudence and interpretation of laws is affecting practice in Germany (Parliament, 2009), Ireland and Italy. Allowed indications for PGD vary also in other countries to a great extent. The diversity of regulation maintains the need for CBRC (Shenfield et al., 2010) and is pertinent also with regards to the application of patient rights in cross-border healthcare (EC-DIR24, 2011).

EC Directive 98/79/EC (EC-DIR98-IVD, 1998) on IVDD (*in vitro* diagnostic devices) is applicable also to genetic tests, but not to in-house assays developed and used in the same facility. The Directive is currently under revision, and it is anticipated that DTC genetic tests will be raised to a higher risk category meaning that they have to meet more stringent criteria in the future.

Recently attention has been drawn to the ethical aspects of preconceptional and prenatal genomic testing in many countries, such as by the Health Council of the Netherlands (Dondorp and de Wert, 2013) and the Nordic Committee on Bioethics (The Nordic-Bioethics, 2010). The main worries relate to the lack of clinical application (within the Analytic validity, Clinical validity, Clinical utility and associated Ethical, legal and social implications framework for evaluation of genetic tests; ACCE framework (Haddow, 2003; USA, 2013) for the majority of tests and the fact that patients/consumers may not receive balanced pre-test information.

Europe is still fragmented with regards to regulation on ART. The rulings of ECHR (European Convention on Human Rights) may in time affect somewhat the states' margin of appreciation. Until people can access services in their own countries, CBRC offers a choice, at least for motivated well-informed patients with the stamina and the necessary means.

Conclusions

The interface between ART and genetics has become more entwined as we increase our understanding about the genetics of infertility and we are able to perform more comprehensive genetic testing. This continually evolving field requires communication between the clinical genetics, IVF teams and patients to ensure that they are fully informed and can make well-considered choices. The genetic basis of male and female infertility will help diagnose the cause of infertility. Also against the background of reports about possible subtle health effects that may be related to epigenetic modifications, there is a growing awareness that

the introduction of new reproductive technologies and treatments needs to be based on sound preclinical and clinical research aimed at collecting evidence about their efficacy and (long-term) safety, as well as their cost-effectiveness. Comprehensive genetic testing of the embryo before implantation raises complex clinical and ethical issues. Couples may increasingly undergo a whole genome scan prior to an IVF (or natural) cycle, and if any serious risk is detected, they can decide which reproductive option would suit them best. The possibility of performing a whole genome scan for PGD is around the corner and would also allow for the detection of *de novo* mutations. As IVF clinics gain higher success rates, and genetic diagnosis helps the treatment of infertile couples, there will need to be much discussion regarding which procedures are clinically and ethically acceptable and how these are regulated. Through these discussions, we must develop sound international policies, and facilitate harmonization of legislation and regulatory practices, including equal access to medically assisted reproduction in Europe, and beyond.

Acknowledgements

M.M. was supported by the Czech Ministry of Health IGA NT/13770-4. J.S. research is partly supported by PEst-C/SAU/LA0002/2011 from FCT, Portuguese Ministry of Education and Science.

Authors' roles

J.H. and M.M. conceived the idea, designed the paper and edited the original paper (Harper *et al.* 2013 which was published as "open access") to produce this document. All authors wrote sections on their topic in the full manuscript (Harper *et al.* 2013 which was published as "open access").

Funding

Participation in the meeting from which this article arose was funded by ESHRE, ESHG and the EuroGentest2 European Union Coordination Action project (FP7 – HEALTH-F4-2010-26146).

Conflict of interest

G.H. currently works for BlueGnome, United Kingdom (an Illumina company) (www.cambridgebluegnome.com). No other authors have a conflict of interest that could influence the content or processing of this manuscript.

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